BOSTER BIOLOGICAL TECHNOLOGY
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Anti-CD4 Antibody Picoband®

Catalog Number: A00344-2

About Cd4

This gene encodes a membrane glycoprotein of T lymphocytes that interacts with major histocompatibility complex class II antigenes and is also a receptor for the human immunodeficiency virus. This gene is expressed not only in T lymphocytes, but also in B cells, macrophages, and granulocytes. It is also expressed in specific regions of the brain. The protein functions to initiate or augment the early phase of T-cell activation, and may function as an important mediator of indirect neuronal damage in infectious and immune-mediated diseases of the central nervous system. Multiple alternatively spliced transcript variants encoding different isoforms have been identified in this gene.

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Overview

Product Name	Anti-CD4 Antibody Picoband®
Reactive Species	Mouse
Description	Boster Bio Anti-CD4 Antibody Picoband® catalog # A00344-2. Tested in ELISA, IHC, WB applications. This antibody reacts with Mouse. The brand Picoband indicates this is a premium antibody that guarantees superior quality, high affinity, and strong signals with minimal background in Western blot applications. Only our best-performing antibodies are designated as Picoband, ensuring unmatched performance.
Application	ELISA, IHC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na2HPO4.
Storage Instructions	Store at -20˚C for one year from date of receipt. After reconstitution, at 4˚C for one month. It can also be aliquotted and stored frozen at -20˚C for six months. Avoid repeated freeze-thaw cycles.
Host	Rabbit
Uniprot ID	P05540

Technical Details

Immunogen	E.coli-derived rat CD4 recombinant protein (Position: K28-I457).
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Rabbit IgG (EK1002) for Western

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	blot, and HRP Conjugated anti-Rabbit IgG Super Vision Assay Kit (SV0002-1) for IHC(P).
Cross Reactivity	No cross-reactivity with other proteins.
Isotype	Rabbit IgG
Form	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.
Purification	Immunogen affinity purified.
Suggested Dilutions	Western blot, 0.25-0.5ug/ml, Mouse Immunohistochemistry (Paraffin-embedded Section), 2-5ug/ml, Mouse ELISA, 0.1-0.5ug/ml,-

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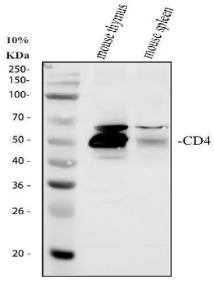
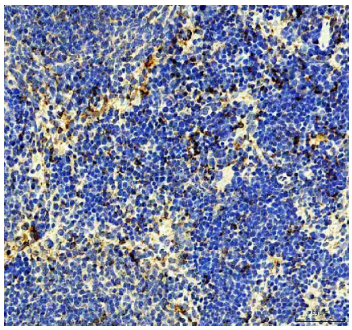
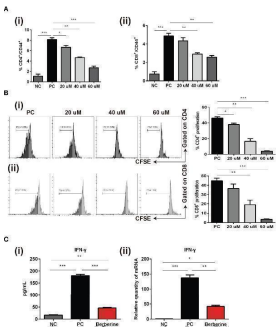
Anti-CD4 Antibody Picoband® (A00344-2) Images



Western blot analysis of CD4 using anti-CD4 antibody (A00344-2). Electrophoresis was performed on a 10% SDS-PAGE gel at 80V (Stacking gel) / 120V (Resolving gel) for 2 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions. Lane 1: mouse thymus tissue lysates, Lane 2: mouse spleen tissue lysates. After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-CD4 antigen affinity purified polyclonal antibody (A00344-2) at 0.5 ug/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody (Catalog # BA1054) at a dilution of 1:5000 for 1.5 hour at RT. The signal is developed using an ECL Plus Western Blotting Substrate (Catalog # AR1196-200) with Tanon 5200 system. A specific band was detected for CD4 at approximately 51 kDa. The expected band size for CD4 is at 51 kDa.

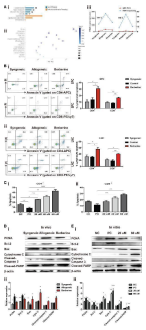


IHC analysis of CD4 using anti-CD4 antibody (A00344-2). CD4 was detected in a paraffin-embedded section of mouse spleen tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-CD4 Antibody (A00344-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

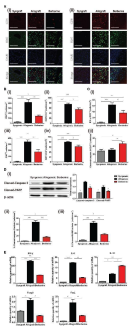


The effects of berberine on CD4 + and CD8 + T cell apoptosis, activation, and proliferation in vitro . (A) T cell activation assay. The percentages of (i) CD4 + CD44 + and (ii) CD8 + CD44 + T cells were determined by flow cytometry. (B) T cell proliferation assay. T cells from naïve C57BL/6 mice were labeled with CFSE and then co-stimulated with anti-CD3/CD28 Abs in the absence or presence of berberine. After 3 days of co-culture, (i) CD4 + and (ii) CD8 + T cell division was determined by flow cytometry. (C) (i) Supernatant levels of IFN-gamma measured by ELISA, and (ii) mRNA expression of IFN-gamma in T cells measured by qPCR. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$ compared to the (Positive Control) PC group.Index in PubMed under a CC BY license. PMID: 33732240

Berberine induces T cell apoptosis via the mitochondrial apoptosis pathway. (A) (i) KEGG functional categories of differentially expressed genes following berberine and saline



treatment. The Y-axis represents the KEGG functional categories. (ii) KEGG analysis of the significantly altered signaling pathways in cell growth- and death-associated genes. The X-axis represents the rich ratio of the number of differentially expressed genes and the Y-axis represents the KEGG pathways. (iii) qPCR analysis of Bcl-2 and TNF-alpha mRNA expression in SPCs collected from heart transplant recipients treated with berberine or not. (B) T cell apoptosis assays in vivo . (i) SPCs and (ii) LNCs were collected at POD 7. The percentages of apoptotic CD4 + and CD8 + T cells were determined by flow cytometry (n = 3 mice/group). (C) T cell apoptosis assay in vitro . T cells from naïve C57BL/6 mice were co-stimulated with anti-CD3/CD28 Abs in the absence or presence of berberine. The percentages of apoptotic (i) CD4 + and (ii) CD8 + T cells were determined by flow cytometry. (D) Berberine activates the mitochondrial apoptosis pathway in vivo . (i) Relative protein expression of Bcl-2, Bax, cytochrome c, cleaved-caspase-3, and cleaved-PARP in SPC. (ii) beta-actin was used as a loading control (n = 3 mice/group), and OD values (relative to beta-actin) are presented as means ± SEMs. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$ compared to the normal saline-treated group. (E) Berberine activates the mitochondrial apoptosis pathway in vitro . Relative protein expression of Bcl-2, Bax, cytochrome c, cleaved-caspase-3, and cleaved-PARP expression in CD3 + T cells. (ii) beta-actin was used as a loading control; OD values (relative to beta-actin) are presented as means ± SEMs. SPCs, spleen cells; LNCs, lymph node cells; POD, post-operative day. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$ compared to the PC group.Index in PubMed under a CC BY license. PMID: 33732240



Phenotypic and functional characteristics of allograft-infiltrating CD4 + or CD8 + T cells. Allografts were recovered at POD 7, and POD 100 syngeneic grafts are shown for comparison. (A) (i) Immunofluorescent staining of CD4 (red), KI67 (green), and 4′,6-diamidino-2-phenylindole (DAPI, blue) in grafts. (ii) Immunofluorescent staining of CD8 (red), KI67 (green), and DAPI in grafts (Scale bar = 200 um; original magnification: ×200). (B) Proportion and absolute number of graft-infiltrating (i) CD4 + T cells and their expression of (ii) KI67, and proportion and absolute number of graft-infiltrating (iii) CD8 + T cells and their expression of (iv) KI67 (n = 3 mice/group). (C) Proportion of (i) IFN-gamma and (ii) cleaved-caspase-3 in graft-infiltrating CD3 + T cells (n = 3 mice/group). (D) (i) Cleaved-caspase-3 and cleaved-PARP protein expression in grafts. Myocardial cell apoptosis co-immunofluorescence staining and expression of (ii) cleaved-caspase-3 and (iii) cleaved-PARP (n = 3 mice/group). (E) Relative mRNA expression of IFN-gamma , IL-6, IL-10 , Foxp3 , and FasL in grafts measured by qPCR (n = 3 mice/group). SPCs, spleen cells; LNCs, lymph node cells; POD, post-operative day. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$ compared to the normal saline-treated group.Index in PubMed under a CC BY license. PMID: 33732240

Effect of berberine treatment on the T cell immune responses to cardiac allografts. (A) MLR responses. Recipient

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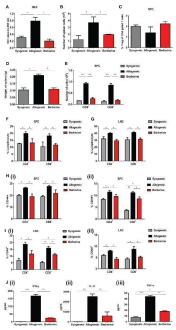

SPCs were isolated at POD 7 (responders) and mitomycin C-treated naïve BALB/c SPCs (stimulators) were co-cultured for 3 days (n = 3 mice/group). Total SPCs were isolated at PDO 7, and the absolute numbers of (B) SPCs were determined by flow cytometry. (C) The percentage of CD4 + Foxp3 + Treg cells was determined by flow cytometry. Syngeneic recipients are shown for comparison (n = 3 mice/group). (D) The spleens of recipient mice were harvested and weighed at POD 7 (n = 5 mice/group). (E) The absolute numbers of CD4 + and CD8 + T cells SPCs (n = 3 mice/group). (F) SPCs and (G) LNCs were isolated at POD 7, and the percentage of CD4 + and CD8 + T cells was determined by flow cytometry. Syngeneic recipients are shown for comparison (n = 3 mice/group). (H) CD44 + CD69 + T cell activation assays. SPCs were isolated at POD 7, and the percentages of (i) CD4 + CD44 + CD69 + and (ii) CD8 + CD44 + CD69 + T cells were determined by flow cytometry (n = 3 mice/group). (I) LNCs were isolated at POD 7, and the percentages of (i) CD4 + CD44 + CD69 + and (ii) CD8 + CD44 + CD69 + T cells were determined by flow cytometry (n = 3 mice/group). (J) Serum plasma levels of proinflammatory cytokines. Peripheral blood was collected at POD 7, and (i) IFN-gamma, (ii) IL-6, and (iii) TNF-alpha plasma levels were measured by ELISA (n = 3 mice/group). SPCs, spleen cells; LNCs, lymph node cells; MLR, mixed lymphocyte reaction; POD, post-operative day. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$ compared to the normal saline-treated group.Index in PubMed under a CC BY license. PMID: 33732240

4 Publications Citing This Product

1. PubMed ID: 10.1177/09636897211054503, Leflunomide Inhibits rat-to-Mouse Cardiac Xenograft Rejection by Suppressing Adaptive Immune Cell Response and NF-kappaB Signaling Activation:

2. PubMed ID: 10.3109/00016489.2011.552525, High dose combination pertussis toxin induces autoimmune inner ear disease in Sprague-Dawley rats

3. PubMed ID: 10.3389/fimmu.2021.616074, Berberine Prolongs Mouse Heart Allograft Survival by Activating T Cell Apoptosis via the Mitochondrial Pathway

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